

about 10 to about 100 microns. Likewise, the length of the sample channel **14** may also vary. For example, the sample channel **14** may have a length that is from about 1 millimeter to about 50 centimeters, and in some embodiments, from about 5 millimeters to about 50 millimeters.

[0040] Printing techniques are generally utilized in the present invention to apply the sample channel **14** due to their practical and cost-saving benefits. For instance, several suitable printing techniques are described in U.S. Pat. No. 5,512,131 to Kumar, et al.; U.S. Pat. No. 5,922,550 to Everhart, et al.; U.S. Pat. No. 6,294,392 to Kuhr, et al.; U.S. Pat. No. 6,509,085 to Kennedy; and U.S. Pat. No. 6,573,040 to Everhart, et al., which are incorporated herein in their entirety by reference thereto for all purposes. For example, in one embodiment, "stamp printing" is utilized to apply the sample channel **14**. Some suitable stamp printing techniques are described in U.S. Pat. No. 5,512,131 to Kumar, et al. and U.S. Pat. No. 5,922,550 to Everhart, et al. For example, an elastomeric stamp may be used to transfer the ink to the substrate surface through contact. The stamp is fabricated by casting polydimethylsiloxane (PDMS) on a master having the inverse of the desired print pattern, which will thereby result in the desired channel pattern. Masters are prepared using standard photolithographic techniques, or constructed from existing materials having microscale surface features. In one embodiment, a photolithographically-produced master is placed in a glass or plastic Petri dish, and a mixture of SYLGARD® silicone elastomer **184** and SYLGARD® silicone elastomer **184** curing agent (Dow Corning Corporation) is poured over it. The polydimethylsiloxane (PDMS) elastomer is allowed to sit at room temperature and is then cured; alternately, for faster curing, the elastomer may be cured at a temperature of from about 60 to about 65° C. When cured, PDMS is sufficiently elastomeric to allow good conformal contact of the stamp and the surface of the substrate **40**.

[0041] The resulting elastomeric stamp is "inked" by exposing the stamp to a solution of the desired material used to help form the fluidic channel. This is typically done by placing the stamp face down in the solution for about 10 seconds to about 10 minutes. The stamp is allowed to dry, either under ambient conditions or by exposure to a stream of air or nitrogen gas. Following inking, the stamp is applied to the surface of the substrate **40**. Light pressure is used to ensure complete contact between the stamp and the substrate **40**. After about 1 second to about 5 minutes, the stamp is then gently peeled from the substrate **40**. Following removal of the stamp, the substrate **40** may be rinsed and dried.

[0042] Stamp printing, such as described above, may be used to prepare channels in various ways. In one embodiment, for example, the elastomeric stamp is inked with a material that significantly alters the surface energy of the substrate so that it may be selectively "wetable" to the monomer or pre-polymer (if post-cured), or polymer used to make the channel. The stamp could have raised features to print the desired channel pattern. An exemplary stamp printing method may involve inking the stamp with a wetting agent, such as hydrophilic self-assembling monolayers (SAMs), including those that are carboxy-terminated. Various examples of such self-assembling monolayers are described in U.S. Pat. No. 5,922,550 to Everhart, et al. In another embodiment, hydrophobic wetting agents may be utilized. Specifically, the inverse of the desired pattern is

stamp printed onto a hydrophilic substrate. Upon exposure of the monomer or pre-polymer (if post-cured), or polymer, the inks would selectively wet only on the substrate **40**, thereby resulting in the desired channel pattern. Another stamp printing technique might simply involve inking an elastomeric stamp with a solution of the monomer or pre-polymer (if post-cured), or polymer. The stamp may have raised features to match the desired channel pattern so that a direct transfer of the channel-forming material would occur on the substrate **40**.

[0043] Still another suitable contact printing technique that may be utilized in the present invention is "screen printing." Screen printing is performed manually or photo-mechanically. The screens may include a silk or nylon fabric mesh with, for instance, from about 40 to about 120 openings per lineal centimeter. The screen material is attached to a frame and stretched to provide a smooth surface. The stencil is applied to the bottom side of the screen, i.e., the side in contact with the substrate upon which the fluidic channels are to be printed. The print material is painted onto the screen, and transferred by rubbing the screen (which is in contact with the substrate) with a squeegee.

[0044] In addition to contact printing, any of a variety of well-known non-contact printing techniques may also be employed in the present invention. In one embodiment, for example, ink-jet printing may be employed. Ink-jet printing is a non-contact printing technique that involves forcing ink through a tiny nozzle (or a series of nozzles) to form droplets that are directed toward the substrate. Two techniques are generally utilized, i.e., "DOD" (Drop-On-Demand) or "continuous" ink-jet printing. In continuous systems, ink is emitted in a continuous stream under pressure through at least one orifice or nozzle. The stream is perturbed by a pressurization actuator to break the stream into droplets at a fixed distance from the orifice. DOD systems, on the other hand, use a pressurization actuator at each orifice to break the ink into droplets. The pressurization actuator in each system may be a piezoelectric crystal, an acoustic device, a thermal device, etc. The selection of the type of ink jet system varies on the type of material to be printed from the print head. For example, conductive materials are sometimes required for continuous systems because the droplets are deflected electrostatically. Thus, when the sample channel is formed from a dielectric material, DOD printing techniques may be more desirable.

[0045] In addition to the printing techniques mentioned above, any other suitable printing technique may be used in the present invention. For example, other suitable printing techniques may include, but not limited to, such as laser printing, thermal ribbon printing, piston printing, spray printing, flexographic printing, gravure printing, etc. Such techniques are well known to those skilled in the art.

[0046] Besides the sample channel **14**, the substrate **80** may also include other channels that serve a variety of purposes. For example, the substrate **80** may include a washing channel (not shown) that provides for the flow of a washing reagent to the detection working electrode **42** to remove any redox labels (described below) that remain unbound. Examples of washing agents may include, for instance, water, a buffer solution, such as PBS buffer, HEPES buffer, etc., and so forth. In addition, a reagent channel (not shown) may also be provided through which